Developing Planetary Protection Technology: Microbial Diversity of the Mars Orbiter "Odyssey" and the Spacecraft Assembly and Encapsulation Facility II. **Myron T. La Duc**, Fei Chen, Roger Kern, Robert Koukol, Amy Baker, and Kasthuri Venkateswaran. California Institute of Technology, Jet Propulsion Laboratory, Planetary Protection Technologies Group, Pasadena, CA 91109.

Sampling the surfaces of both spacecraft and their clean-room assembly facilities is crucial in monitoring the microbial burden associated with these pseudo-sterile, oligotrophic environments. Here, we present the results of a study in which several surface samples, retrieved from both the Mars Odyssey Spacecraft and the Kennedy Space Center (KSC) Spacecraft Assembly and Encapsulation Facility II (SAEF-II), were processed and evaluated by both molecular and traditional culture-based methods for the microbial diversity. The findings of this study improve our current understanding of the microbial community structure, diversity, and dispersal in a spacecraft assembly facility, as well as physically associated with co-located spacecraft. Surfaces of 25 cm^2 (spacecraft) or 1 m^2 (SAEF-II) were swabbed or wiped, respectively, and were examined for total heterotrophic aerobes and spore-formers. Samples were further subjected to nucleic acid extraction, and 16S rDNA fragments were PCR amplified with eubacterial biased universal primers and cloned. Approximately 30 isolates grown by traditional culture-based techniques were included for 16S rDNA sequencing. For the most part, the population dynamics remained consistent when compared between the spacecraft and assembly facility libraries. Predominant microbes, as indicated by molecular methods, included members of the genera *Variovorax* and *Aquaspirillum*. Members of the *Mesorhizobium, Bradyrhizobium*, *Enterococcus*, *Ralstonia,* and *Bacillus* genera were also found to span the various libraries but in less abundance. Traditional culture-based techniques validated the presence of *Bacillus* and *Ralstonia*, while illuminating a larger diversity in revealing the presence of *Staphylococcus*, *Comamonas, Microbacterium*, and Actinomycetales. The bulk of these findings make sense, since species of *Ralstonia*, *Rhizobium*, *Variovorax*, and *Bacillus* are known to frequently inhabit rhizospheric environments, like that surrounding the KSC facility, and can thus easily gain way into the clean-room via mixing of surrounding air upon human entry and exit. Also, *Aquaspirillum* species are known to inhabit freshwater and brackish ponds, much like the ones at KSC. Of particular interest to the authors was the presence of *Nicotiana tabacum* chloroplast 16S rDNA in one of the spacecraft samples. The lack of tobacco farming anywhere in the vicinity of the KSC facility leads the authors to speculate that this contamination arose from human contact with the spacecraft, specifically after handling a cigarette, cigar, or other tobacco products. Overall, our findings validate the purpose of planetary protection activities, which improve our knowledge of the types of microbial burden present, and their methods of entry into spacecraft assembly facilities.